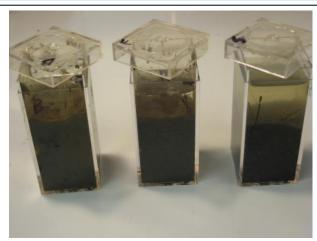
Science Lesson Plan:

Oxygen Measurements in a Microbial Mat

Brad Bebout, Allison Pfeifer, Erich Fleming, Adrienne Frisbee, Jan Winter NASA Ames Research Center

Background

Microbes have shaped the atmosphere and climate of Earth over geologic time. Microbes may be important in the search for life both within our solar system and on extrasolar planets. Some of the earliest and most widespread evidence of life on Earth were in the form of microbial mats. Microbial mats are complex, usually layered collections of microorganisms that can be observed in a number of aquatic ecosystems. They are complete ecosystems in miniature. In this activity, students use microbial mats that they have created to investigate biogeochemical cycling of oxygen.



Living microbial mats constructed in plastic boxes.

Main Concepts

Photosynthetic microbial mats constructed in the laboratory (see "How to Make a Microbial Mat" activity) can be used for classroom investigations of photosynthesis, respiration, and other biogeochemical processes.

Scientific Question

How do microbial mats react to changes in environmental conditions?

Objectives

- 1) Students will take measurements of both oxygen production using Vernier probes and growth of their microbial mat in millimeters over four consecutive weeks.
- 2) Students see comparisons between the overall growth of their microbial mat to algae (high oxygen producer) and to soil (low oxygen producer).
- 3) Students will understand that the greater microbial mat growth that occurs, the more oxygen will be produced as a by-product of photosynthetic cyanobacteria (top layer microbes). (After full maturity; however, oxygen production may begin to decrease, as the mat becomes a closed ecosystem.)
- 4) Students will identify microbes using a microscope and determine how each microbe functions as part of the entire microbial mat ecosystem.

Abstract of Lesson

Microbial mats are defined as layered communities of microorganisms that are only a few millimeters thick. Many biogeochemical processes are conducted by the organisms in the mat to sustain life. For example, the mats perform photosynthesis, and respiration, and can acquire nitrogen through the process of nitrogen fixation. The mat community is a fully functioning ecosystem containing both producers and consumers or organic carbon. The upper layers of the mat contain cyanobacteria that create their own food (organic carbon) through the process of photosynthesis. Light energy is used to split electrons from water and the energy carried by these electrons is used to reduce carbon dioxide into sugars, which are, in turn, used by the cyanobacteria for food. In the process, cyanobacteria in the surface (sunlit) layer also produce oxygen. Below this layer, other bacteria that are tolerant of oxygen, utilize the energy liberated by combining the sugars with oxygen for growth. At the bottom layer of the mat, where oxygen does not penetrate, other microbes, which can combine the organic matter with chemicals other than oxygen, may be found. These microbes utilize a number of chemicals other than oxygen to oxidize the organic matter left behind by oxygen utilizing microbes. These chemicals include nitrate, manganese, iron, and sulfate. Many of these microbes actually cannot tolerate the presence of oxygen.

In this exercise, we will the microbial mats created earlier to look at oxygen production. Although there is a procedure written out below for one investigation of oxygen production, this should be taken as only a starting point. For example, what are the effects of temperature, light intensity, light color, community composition, age of the mats, etc. on oxygen production in the light and consumption in the dark?

Prerequisite Concepts

Students should have general familiarity with laboratory work.

Major Concepts

- 1. Microbial mats are complicated microbial ecosystems.
- 2. Microbial mats display vertical structure (e.g., they have oxygen available at the top of the mat, but not the bottom).
- 3. The microorganisms in mats both orient themselves to vertical gradients and contribute to making the gradients as a result of their metabolism.

Standards Met:

Project 2061 AAAS 1993: 5.D.1

5.F.8

5.F.9

Materials List:

- Vernier probes (dissolved oxygen) with TI calculators and/or LoggerPro software with classroom computers
- 2) Algae sample (about 250mL)
- 3) Soil sample (about 250mL)

- 4) Rulers (mm)
- 5) Microbial Mats (students)
- 6) Microscopes (dissecting microscopes will also work)
- 7) Slides, cover slips, water dropper
- 8) Plastic straw (for removing a column of microbial mat for microscope)
- 9) Student hand-outs: http://microbes.arc.nasa.gov/download/pdf/Strom Expl Plan.pdf
- 10) Glossary and pictures of microbes (from above website) for use in identifying species under the microscope as well as help with answering questions on the student handout

Lab and Field Safety

Microbes and microbial mat communities consist of a combination of harmless bacteria and algae. However, when sampling microbial mats from the outdoor environment, you should always consider the possibility that non-point source pollution could occur in that area. Students should always wash their hands after handling microbial mats or "mud." Never eat during the lab. Goggles should also be worn during the lab.

Procedure

Teacher Setup: Two days prior to lab day:

1) Prepare a class demonstration using algae and potting soil sample. You can get algae easily from any local pond or you can order algae from a biological supply company and grow it in media (allow for more time if ordering). Place about 250mL of algae in a plastic container (if you use the 250 mL Nalgene Bottle w/Lid sold by Vernier, it will interface perfectly with their gas sensor) and completely cover with water until you create a 1-2 inch water column either above or below algae (some algae will float). Allowing algae to sit in the water for at least two days prior to the start of the lab. This will allow time for oxygen produced by the algae to dissolve into the water column surrounding the algae. In a separate container, use potting soil and cover with water until you create a 1-2 inch water column above the soil. You may need to allow time for the soil to settle to the bottom of you container in order to determine if you actually have 1-2 inches of water above the soil. You can also use soil or mud from a garden as long as you are not mixing it with anything "green" (ex. no plants, moss, algae, etc).

Teacher Setup: One day prior to lab day:

1) Place microscope, cover slips, water dropper, pictures of microbes (for viewing microbes found within student made microbial mats) and one plastic straw per student at each lab table. Also, gloves and goggles are required for each student. Note: Students may need to be shown how to prepare a wet-mount slide.

Lab day:

 Demo: Test the dissolved oxygen of algae and soil in water using Vernier probes (http://www.vernier.com/). This demonstration will serve as a reference of comparison to the amount of dissolved oxygen generated in student-made microbial mats. After taking the reading of algae and soil, you should get a very high reading for algae (photosynthesis) and very

- low or negative value reading for soil. Refer to "Vernier Probe Use" for more detailed directions on how to use Vernier probes to test for dissolved oxygen.
- 2) Allow time for students to record these two readings onto their papers (refer to attachment: "Making a Microbial Mat-Student).
- 3) Allow students to begin testing the dissolved oxygen. This will serve as the reading for week 1. Students will record reading in the data table located on their handout. Students will continue to take readings of their growing microbial mats once a week for a total of four consecutive weeks. If time is limited, it is not necessary to repeat the algae and soil demonstration as they can continue to use the original readings (from week 1).
- 4) Students should make the connection that it is the photosynthetic microbes, such as the cyanobacteria, found at the top of the microbial mat that is allowing oxygen to be dissolved in the water column directly above the mat.
- 5) Note: students can share probes while other students view microbes under the microscope. Students will get a number that will indicate the amount of dissolved oxygen given off by the microbial mat.
- 6) Directions for viewing microbes under the microscope: Students can use the plastic straw by inserting it vertically into their mat without damaging the entire mat. While the straw in inside the mat, place one finger on top of the straw to cover the hole, remove straw from the mat by gently lifting up on the straw. Once the straw is removed, students can remove the contents inside the straw by gently squeezing the straw and pushing down the length of the straw so that the contents are squeezed out of the straw.

Vernier Probe Use:

- 1. Most Vernier kits will come with directions on how to sample for Dissolved Oxygen. Here is an example using TI-84 Plus calculators:
 - a. First student using probe: Plug in Easy Link cable into the USB of calculator (the calculator will automatically turn on)
 - b. Plug in Oxygen probe into the Easy Link.
 - c. IMPORTANT: Put the probe in regular tap water with only the tip covered for 10 minutes after turning on the calculator. Remove from water before going on.
 - d. Go to File, New-Press Enter
 - e. Go to Setup, Single Point-Press Enter (now the probe is ready to test the dissolved oxygen of the sample water).
 - f. Put the end of the sensor into the sample water directly above the top of the microbial mat (sensor must be in water). Get as close as you can to the top of the mat without pushing the sensor into the mat.
 - g. IMPORTANT: You must gently stir the sensor while the reading is being taken by moving the probe around in a slow circle.
 - h. Press start. After 10 seconds, make sure it says Event number 1. If it has any other number (for the first person), go back to step g.
 - i. Record the number that appears in the top write corner of the screen onto their papers.
 - j. Press OK.
 - k. Remove sensor from mat sample.
 - I. Repeat steps f-k for each student